

WHAT IS CLAIMED IS:

1. An isolated nucleic acid encoding the GEF-H1b polypeptide of SEQ ID NO:2.
2. An isolated nucleic acid comprising the sequence of SEQ ID NO:1.
3. An isolated GEF-H1b polypeptide comprising the sequence of SEQ ID NO:2.
4. An isolated GEF-H1/PAK4complex.
5. The isolated GEF-H1/PAK4complex of claim 4, wherein said GEF-H1 is GEF-H1b.
6. The isolated GEF-H1/PAK4complex of claim 5, wherein said GEF-H1b comprises the sequence of SEQ ID NO:2.
7. An isolated nucleic acid encoding the GEF-H1 peptide of SEQ ID NO:3.
8. A peptide consisting essentially of the sequence of SEQ ID NO. 3.
9. An isolated nucleic acid encoding the GEF-H1 peptide of SEQ ID NO:4.
10. A peptide consisting essentially of the sequence of SEQ ID NO. 4.
11. The isolated GEF-H1b polypeptide of claim 3 lacking the amino acid residues between 162 and 354 of SEQ ID NO. 2.

12. The peptide of any one of claims 3, 8, or 10 that comprises at least one phosphorylated amino acid.

13. A method for detecting PAK4 activity in a sample, comprising detecting the presence or level of phosphorylated GEF-H1, wherein the detection of phosphorylated GEF-H1 indicates the presence of at least one active PAK4.

14. The method of claim 13, wherein said GEF-H1 comprises the sequence described in any one of SEQ ID NOs. 2, 3 or 4.

15. The method of claim 13, wherein said PAK4 is in a cell.

16. The method of claim 15, wherein said cell is a tumor cell.

17. The method of claim 16, wherein said tumor cell is in a mammal.

18. The method of claim 17, wherein said mammal is selected from the group consisting of a human, rat, mouse, dog, rabbit, pig, sheep, cow, horse, cat, primate, goat, or monkey.

19. A method of identifying a substance that modulates the interaction between PAK4 and GEF-H1 polypeptide comprising: (a) exposing GEF-H1 polypeptide to a candidate substance to form a mixture and then (b) introducing into said mixture a PAK4 enzyme; and (c) measuring the amount of GEF-H1 polypeptide phosphorylated before and after exposing said GEF-H1 polypeptide to said candidate substance, wherein a decrease or increase in the amount of phosphorylated GEF-H1 polypeptide after exposure to said candidate substance indicates that said candidate substance is a substance that modulates the interaction between PAK4 and GEF-H1 polypeptide.

20. The method of claim 18, wherein said GEF-H1 polypeptide has the sequence of any one of SEQ ID NO. 2, SEQ ID NO. 3 or SEQ ID NO. 4.

21. A GEF-H1-specific antibody directed against a peptide comprising the sequence described in SEQ ID NO. 3.

22. A GEF-H1-specific antibody directed against a peptide comprising the sequence described in SEQ ID NO. 4.

23. A GEF-H1-specific antibody directed against a peptide comprising the sequence described in SEQ ID NO. 3, wherein said peptide comprises a phosphorylated serine.

24. The GEF-H1-specific antibody of claim 23, wherein said serine is serine-810.

25. A GEF-H1-specific antibody directed against a peptide comprising the sequence described in SEQ ID NO. 4, wherein said peptide comprises a phosphorylated serine.

26. The GEF-H1-specific antibody of claim 25, wherein said serine is serine-67.

27. A method of identifying a substance that modulates the interaction between PAK4 and GEF-H1 polypeptide comprising: (a) exposing PAK4 to a candidate substance to form a mixture and then (b) introducing into said mixture a GEF-H1 polypeptide; and (c) measuring the amount of GEF-H1 polypeptide phosphorylated, wherein a decrease or increase in the amount of phosphorylated GEF-H1 polypeptide in comparison to a control in which PAK4 is not exposed to said candidate substance indicates that said candidate substance is a substance that modulates the interaction between PAK4 and GEF-H1 polypeptide.

28. The method of claim 27, wherein said GEF-H1 polypeptide has the sequence of any one of SEQ ID NO. 2, SEQ ID NO. 3 or SEQ ID NO. 4.

29. The method of claims 19 or 27, wherein the step of determining whether GEF-H1 is phosphorylated in step (c) is performed by using a GEF-H1-specific antibody directed against a peptide comprising at least one of phosphorylated serine-810 of SEQ ID NO. 2 or phosphorylated serine-67 of SEQ ID NO. 2 to detect phosphorylated GEF-H1b.

30. The method of claims 19 or 27, wherein said candidate substance causes a decrease in total GEF-H1 phosphorylation.

31. A method for identifying a substance that modulates the interaction between PAK4 and GEF-H1b comprising (i) contacting a candidate substance with a GEF-H1b-PAK4 complex and then (ii) determining whether said compound disrupts said complex.

32. A method for determining whether a candidate substance inhibits PAK4 kinase activity in a mammal, comprising (i) measuring in a mammal the level of phosphorylation of a GEF-H1 protein comprising the sequence of SEQ ID NO. 2; (ii) exposing said mammal to a candidate substance; and then (iii) measuring in the mammal the level of phosphorylation of said GEF-H1 protein, wherein a decrease in the level of phosphorylation of said GEF-H1 protein in step (iii) relative to the level measured in step (i) indicates that said candidate substance is an inhibitor of PAK4 kinase.

33. The method of claim 32, wherein said mammal is a human, rat, mouse, dog, rabbit, pig, sheep, cow, horse, cat, primate, goat, or monkey.

34. The method of claim 32, wherein said mammal is a human.

35. A method for treating cancer in a mammal, comprising administering a substance identified by any one of claims 19, 27, 30 or 32, to said mammal.

36. A method for determining the presence of activated PAK4 in a cell sample comprising (i) obtaining a cellular lysate from a cell sample; (ii) isolating and/or separating proteins from the cell lysate preparation; and (iii) detecting the presence of GEF-H1b phosphovariants, wherein detecting that serine-810 or serine-67 is phosphorylated indicates that said cell sample comprises activated PAK4.

37. A method for screening for a drug that inhibits PAK4 kinase activity comprising (i) obtaining a cellular lysate from a cell sample, (ii) applying to said lysate a candidate drug; (iii) isolating and/or separating proteins from said cell lysate preparation; and (iv) detecting the presence of GEF-H1b phosphovariants, wherein detecting that serine-810 or serine-67 is phosphorylated indicates that said drug does not inhibit PAK4 kinase activity.

38. The method of either claim 36 or 37, wherein said cell sample is preferably a tumor cell sample.

39. A peptide comprising at least one of SEQ ID NO. 6 or SEQ ID NO. 20.

40. A peptide consisting essentially of at least one of SEQ ID NO. 6 or SEQ ID NO. 20.

41. A peptide of less than 20 amino acids comprising the sequence of SEQ ID NO. 6.

42. A peptide of less than 25 amino acids comprising the sequence of SEQ ID NO. 6.

43. A peptide of less than 30 amino acids comprising the sequence of SEQ ID NO. 6.

44. A peptide of less than 20 amino acids comprising the sequence of SEQ ID NO. 20.

45. A peptide of less than 25 amino acids comprising the sequence of SEQ ID NO. 20.

46. A peptide of less than 30 amino acids comprising the sequence of SEQ ID NO. 20.

47. A method for detecting cell proliferation, cell motility and/or cell invasion in a mammal comprising monitoring at timepoint A and at least one other timepoint, B, the phosphorylation level of GEF-H1 in a sample taken from said mammal, wherein an increase in the phosphorylation level of GEF-H1 in said sample between said timepoints, is indicative of cell proliferation, cell motility and/or cell invasion.

48. The method of claim 47, wherein said GEF-H1 is a GEF-H1b protein comprising the sequence of SEQ ID NO. 2.

49. The method of claim 47, wherein said sample is a sample of said mammal's skin, blood, or cells.

50. The method of claim 49, wherein said cells are tumor cells.

51. The method of claim 47, wherein said mammal is selected from the group consisting of a human, rat, mouse, dog, rabbit, pig, sheep, cow, horse, cat, primate, goat, or monkey.

52. A polynucleotide comprising the sequence of SEQ ID NO. 1.
53. A polynucleotide encoding the polypeptide described in any one of SEQ ID NOs. 2, 3 or 4.
54. A vector comprising the polynucleotide of either claim 52 or 53.
55. A cell comprising the vector of claim 54.
56. A method for treating cancer in an individual, comprising inhibiting GEF-H1 activity in said individual.
57. The method of claim 56, wherein said GEF-H1 is at least one of GEF-H1a, GEF-H1b or GEF-H1c.
58. The method of claim 57, wherein said GEF-H1b comprises the amino acid sequence described in SEQ ID NO. 2.
59. The method of claim 56, wherein said step of inhibiting GEF-H1 activity comprises inhibiting expression of an endogenous gene encoding said GEF-H1.
60. The method of claim 56, wherein GEF-H1 activity is inhibited in or near a population of cancerous cells present in said individual.
61. A method for reducing cell proliferation and anchorage-independent cell growth in a cell sample, comprising inhibiting GEF-H1 activity in said cell sample.
62. The method of claim 61, wherein said GEF-H1 is at least one of GEF-H1a, GEF-H1b or GEF-H1c.

63. The method of claim 61, wherein said GEF-H1b comprises the amino acid sequence described in SEQ ID NO. 2.

64. An isolated polynucleotide encoding a polypeptide comprising the amino acid sequence of SEQ ID NO. 21, wherein said polypeptide is capable of binding to a guanine-nucleotide exchange factor, and whereupon binding of said polypeptide to said guanine-nucleotide exchange factor prevents or reduces guanine-nucleotide exchange factor phosphorylation by PAK4.

65. The isolated polynucleotide of claim 64, wherein said polypeptide binds to amino acids 763-921 of GEF-H1S.

66. An isolated polynucleotide encoding a polypeptide consisting essentially of the amino acid sequence of SEQ ID NO. 21.

67. An isolated polynucleotide encoding a polypeptide consisting of the amino acid sequence of SEQ ID NO. 21.

68. An isolated polynucleotide encoding a polypeptide that has at least 70% sequence identity to the amino acid sequence of SEQ ID NO. 21, and wherein said polypeptide is capable of binding to a guanine nucleotide-exchange factor.

69. An isolated polynucleotide encoding a polypeptide comprising the amino acid sequence of SEQ ID NO. 22.

70. An isolated polynucleotide encoding a polypeptide consisting essentially of the amino acid sequence of SEQ ID NO. 22.

71. An isolated polynucleotide encoding a polypeptide consisting of the amino acid sequence of SEQ ID NO. 22.

72. An isolated polynucleotide encoding a polypeptide that has at least 90% sequence identity to the amino acid sequence of SEQ ID NO. 22, and wherein said polypeptide is capable of binding to a guanine nucleotide-exchange factor.

73. A polypeptide consisting essentially of the amino acid sequence of SEQ ID NO. 21.

74. The polypeptide of claim 73, wherein said polypeptide comprises a label.

75. The polypeptide of claim 74, wherein said label is selected from the group consisting of a biotin, a HIS-tag, and a radiolabel.

76. A polypeptide consisting essentially of the amino acid sequence of SEQ ID NO. 22.

77. The polypeptide of claim 76, wherein said polypeptide comprises a label.

78. The polypeptide of claim 77, wherein said label is selected from the group consisting of a biotin, a HIS-tag, and a radiolabel.

79. A polypeptide consisting essentially of an amino acid sequence that has at least 70% sequence identity with SEQ ID NO. 21, wherein said polypeptide is capable of binding to a guanine nucleotide-exchange factor.

80. A polypeptide consisting essentially of an amino acid sequence that has at least 76% sequence identity with SEQ ID NO. 21, wherein said polypeptide is capable of binding to a guanine nucleotide-exchange factor.

81. A recombinant polypeptide comprising an amino acid sequence that has at least 70% sequence identity with the amino acid sequence denoted by residues 276-324 of p21-activated-kinase 4, and at least one other polypeptide sequence, wherein said recombinant polypeptide does not comprise the amino acid sequence denoted by residues 1-276 of p21-activated-kinase 4, nor the amino acid sequence denoted by residues 324-985 of p21-activated-kinase 4.

82. The polypeptide of claim 80, wherein said polypeptide comprises a label.

83. The polypeptide of claim 82, wherein said label is selected from the group consisting of a biotin, a HIS-tag, and a radiolabel.

84. The polypeptide of claim 81, wherein said polypeptide comprises a label.

85. The polypeptide of claim 84, wherein said label is selected from the group consisting of a biotin, a HIS-tag, and a radiolabel.

86. A polypeptide consisting essentially of an amino acid sequence that has at least 90% sequence identity with SEQ ID NO. 22, wherein said polypeptide is capable of binding to a guanine nucleotide-exchange factor.

87. A recombinant polypeptide comprising an amino acid sequence that has at least 70% sequence identity with the amino acid sequence denoted by residues 298-324 of p21-activated-kinase 4, and at least one other polypeptide sequence, wherein said recombinant polypeptide does not comprise the amino acid sequence denoted by residues 1-276 of p21-activated-kinase 4, nor the amino acid sequence denoted by residues 324-985 of p21-activated-kinase 4.

88. A polypeptide comprising an amino acid sequence that has at least 70% sequence identity with the amino acid sequence denoted by residues 276-324 of p21-activated-kinase 4, wherein said polypeptide does not comprise the amino acid sequence denoted by residues 1-276 of p21-activated-kinase 4, and does not comprise the amino acid sequence denoted by residues 324-985 of p21-activated-kinase 4.

89. A polypeptide comprising an amino acid sequence that has at least 90% sequence identity with the amino acid sequence denoted by residues 298-324 of p21-activated-kinase 4, wherein said polypeptide does not comprise the amino acid sequence denoted by residues 1-276 of p21-activated-kinase 4, and does not comprise the amino acid sequence denoted by residues 324-985 of p21-activated-kinase 4.

90. A method for detecting the presence of a guanine nucleotide-exchange factor in a biological sample, comprising:

(i) incubating a biological sample with a p21-activated-kinase-derived polypeptide, and

(ii) determining if any of said p21-activated-kinase-derived polypeptide is bound to a guanine nucleotide-exchange factor,

wherein (a) said p21-activated-kinase-derived polypeptide comprises an amino acid sequence that has at least 90% sequence identity with the amino acid sequence denoted by residues 298-324 of p21-activated-kinase 4,

(b) said p21-activated-kinase-derived polypeptide does not comprise the amino acid sequence denoted by residues 1-276 of p21-activated-kinase 4, and

(c) said p21-activated-kinase-derived polypeptide does not comprise the amino acid sequence denoted by residues 324-985 of p21-activated-kinase 4.

91. The polypeptide of claim 90, wherein said p21-activated-kinase-derived polypeptide comprises a label.

92. The polypeptide of claim 91, wherein said label is selected from the group consisting of a biotin, a HIS-tag, and a radiolabel.

93. The method of claim 92, wherein the step of determining if any of said p21-activated-kinase-derived polypeptide is bound to a guanine nucleotide-exchange factor, comprises removing unbound, radiolabeled p21-activated-kinase-derived polypeptide from said biological sample, and then measuring the level of radioactivity in said sample.

94. A pharmaceutical composition comprising a polypeptide that consists essentially of the amino acid sequence of SEQ ID NO. 21.

95. A method of inhibiting PAK4-mediated phosphorylation of a guanine-nucleotide exchange factor comprising exposing a guanine-nucleotide exchange factor to a polypeptide consisting essentially of the amino acid sequence of SEQ ID NO. 21, wherein said polypeptide binds to said guanine-nucleotide exchange factor, thereupon inhibiting PAK4 phosphorylation of said guanine-nucleotide exchange factor.

96. The method of claim 95, wherein said guanine-nucleotide exchange factor is in a biological sample.

97. The method of claim 96, wherein said biological sample is obtained from a mammal.

98. The method of claim 97, wherein said mammal is a human, mouse, rat, pig, cow, dog, cat, horse, or monkey.

99. The method of claim 98, wherein said mammal is a human.

100. The method of claim 95, wherein the step of exposing said guanine-nucleotide exchange factor to said polypeptide comprises administering to a mammal a pharmaceutical composition that comprises said polypeptide.

101. The method of claim 100, wherein said pharmaceutical composition comprising said polypeptide is any one of a tablet, aerosol, powder, liquid, gel, cream, suppository.

102. A method for identifying a molecule, which disrupts an interaction between PAK4 and a guanine-nucleotide exchange factor, comprising: (a) exposing a guanine-nucleotide exchange factor to a polypeptide consisting essentially of the amino acid sequence of SEQ ID NO. 21, wherein said polypeptide binds to said guanine-nucleotide exchange factor to form a complex; (b) adding one or more test molecules to said complex; and (c) determining whether said polypeptide and said guanine-nucleotide exchange factor are dissociated from one another after said test molecule(s) is added to said complex.

103. A method of inhibiting PAK4 phosphorylation of a GEF-H1 isoform, comprising introducing into a cell a vector that comprises a polynucleotide, which encodes the polypeptide of SEQ ID NO. 21, wherein said polynucleotide is expressed to produce said polypeptide, and wherein said polypeptide binds to a GEF-H1 isoform in said cell, thereby inhibiting PAK4 phosphorylation.